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RESEARCH NOTE

First description of DHA-1 *ampC* β -lactamase in *Proteus mirabilis*

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ABSTRACT

This report describes the first occurrence of the DHA-1 *ampC* β -lactamase gene in *Proteus mirabilis*. The organism was isolated from the vaginal flora of a pregnant woman in a French hospital. The DHA-1 β -lactamase gene was identified

on the basis of phenotypic characteristics, PCR amplification and sequencing. Antagonism between cefoxitin and the other cephalosporins suggested inducible production of the DHA-1 enzyme.

Keywords *ampC*, β -lactamase, class C β -lactamases, DHA-1, *Proteus mirabilis*, resistance

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Proteus mirabilis is a Gram-negative bacterium that is naturally sensitive to ampicillin and cephalosporins because of the lack of a chromosomal *ampC* β -lactamase gene. However, this organism can become resistant to ampicillin by acquisition of a TEM-type penicillinase. Inhibitor-resistant TEM enzymes and resistance to broad-spectrum cephalosporins, resulting from the acquisition of TEM-type extended-spectrum β -lactamases, have also been described in *P. mirabilis* [1,2]. Most recently, class C β -lactamases, conferring resistance to 7 α -methoxy-cephalosporins, have appeared in this species [3,4]. The *ampC* genes described in *P. mirabilis* belong to the ACC or CMY families, related to the *Hafnia alvei* and *Citrobacter freundii* *ampC* chromosomal genes, respectively, and have been reported to be either plasmid-encoded or inserted on the chromosome (probably by transposition) in *P. mirabilis* [3–6]. The DHA-1 plasmid-encoded *ampC* gene was first described in *Salmonella enterica* in Saudi Arabia (with recent spread to the UK), and in *Klebsiella pneumoniae* [7–11]. The present report describes the first occurrence of a DHA-1 *ampC* gene in *P. mirabilis*.

The patient concerned was a pregnant woman, aged 34 years, who was admitted to the maternity ward of Robert-Debré Hospital, Paris. A *P. mirabilis* isolate resistant to cephalothin was isolated from a vaginal swab taken after 37 weeks of amenorrhoea. The patient gave birth by Caesarian section after 40 weeks of amenorrhoea to a healthy infant that was not colonised by *P. mirabilis*. Two days after delivery, the patient had a fever (38.6°C) and was treated with cefotaxime for 3 days, but with no documented infection (blood and urine cultures gave negative results). Ten days after delivery, a *P. mirabilis* isolate resistant to cephalo-

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thin, together with *Pseudomonas aeruginosa*, was isolated from a suppuration of the Caesarian scar. Local treatment with antiseptic agents was sufficient to cure this superficial infection, and no antibiotic was given to the patient at that time.

The identification of the organism as *P. mirabilis* was by Gram's stain and tests with the API 20E and ID32GN systems (bioMérieux, Marcy l'Etoile, France). Antimicrobial susceptibility was tested by the NCCLS disk diffusion method on Mueller–Hinton agar (Sanofi-Diagnostics Pasteur, Marnes-La-Coquette, France). MICs of amoxycillin, cefotaxime, ceftazidime, cefepime and imipenem, as determined by Etests (Biodisk, Solna, Sweden), were >256, 0.5, 1.5, 0.064 and 0.25 mg/L, respectively. The isolate was resistant to amoxycillin, cephalothin and amoxycillin–clavulanic acid, but remained sensitive to ticarcillin, cefotaxime, ceftazidime and imipenem. Double-disk diffusion tests did not indicate synergy between clavulanic acid and cephalosporins (cefotaxime, ceftazidime, cefepime), but antagonism between cefoxitin and the other cephalosporins was observed, thus suggesting the inducible production of a group C β -lactamase by this isolate.

Total DNA was extracted with the QIAamp DNA Mini Kit (Qiagen, Courtaboeuf, France). PCRs for CMY-2- and ACC-1-like genes were performed as described previously [3,12], but were negative. The *ampC* and *ampR* genes were amplified with three pairs of primers generating overlapping PCR fragments in a 2243-bp region: DHA upper1 (5'-CTCATCCTCCATAAACAGC-3') and DHA lower1 (5'-TTATCTCACACCTTTAT-TACT-3'); DHA upper2 (5'-AGATACATTGCCATTTCCAG-3') and DHA lower2 (5'-ACT-TGCCGCCGTTACTCACA-3'); and DHA upper3 (5'-TGTGCCATCAGCGTTTATT-3') and DHA lower3 (5'-TGGAAGGTGAGTGAGTTTAA-3'). Sequencing of the PCR products yielded a sequence that was 100% identical to the published sequences of the *bla*_{DHA-1} and *ampR* genes [9]. To our knowledge, this is the first report of a DHA-1 *ampC* β -lactamase gene in *P. mirabilis*.

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